octahedrally coordinated to five nitrogen atoms and a sulfur atom. The iodine atom, which lies on a twofold axis, bridges the two crystallographically related cobalt centers by means of a S–I–S linkage. The presence of five nitrate anions (three of which are disordered) confirms the +1 oxidation state assignment for the iodine atom. The S–I bond length of 2.619 (2) Å is in good agreement with the 2.629 (1) Å reported for $\{[(NH_2)_2C{=}S]_2I\}^+$.[13] The S–I–S linkage is nearly linear (bond angle of 173(1)°), as is the analogous S–Ag–S linkage in $\{[(en)_2Co(SCH_2COO)]_2Ag\}^{3+}$ (169.4(1)°).[18] The S–I–S angle in the bis(thiourea) adduct is required to be 180.0° by symmetry. A linear S–I–S array is expected for a five-electron pair, trigonal-bipyramidal, I^+ center in which the two sulfur atoms occupy axial positions and three lone electron pairs occupy equatorial positions.

The chemistry of $\{[(en)_2Co(SCH_2CH_2NH_2)]_2I\}^{5+}$ as both a prototype coordinated sulfenyl iodide and as a potential aqueous I^+ donor is under active investigation.[19]

Acknowledgment. This work is dedicated to the late Dr. Michael Hill, whose initial experiments helped lead us into the study of coordinated sulfenyl iodides. Financial support by the National Science Foundation, Grant No. CHE 76-19681 and CHE 79-26497 to E.D., is gratefully acknowledged.

Supplementary Material Available: Tables of atomic positional and thermal parameters and figure containing all atom labels (3 pages). Ordering information is given on any current masthead page.

(19) A more complete report of this work will be published later in this journal by the same authors.

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Further Verification of Fluorescence-Detected Circular Dichroism

Sir:

Fluorescence-detected circular dichroism (FDCD) measures the difference in fluorescence intensity excited by left and right circularly polarized light (LCPL and RCPL) and is potentially a powerful probe of conformational changes in macromolecules.[1-4] However, two recent communications have raised questions concerning the interpretation of FDCD in this latter application.[5,6] Moreover, experiments designed to test FDCD theory have been limited by artifacts.[7] We report here experiments in which FDCD gives quantitatively predicted spectra under conditions similar to those encountered with macromolecules. The results indicate that artifacts can be abolished, and theories useful for interpretation of transmission circular dichroism (CD) spectra will also be useful for FDCD.

The first potential problem with FDCD measurements on macromolecules was pointed out by Ehrenberg and Steinberg.[5] They noted that the fluorescence will be polarized, and therefore photoselection effects must be considered. A detailed theory for this case has been derived.[8] One result is that for an excitation beam propagating in the $\hat{k}$ direction, with a photomultiplier oriented perpendicular to this along the $\hat{j}$ axis, with a polarizer in front of it, the measured signal, S_F, is[7,8]

$$S_F = \frac{K(F_L - F_R)}{F_L + F_R} = -14.32\left[\frac{8R(1 + \cos^2\phi) + (8R_{33}/3)(2 - 3\cos^2\phi)}{D(4 - \cos^2\phi) - (D_{33}/3)(2 - 3\cos^2\phi)} - 2R_l\right] \quad (1)$$

K is an instrument constant, F_L and F_R are fluorescence intensities measured for left and right circularly polarized excitation, ϕ is the angle the polarizer axis makes with the $\hat{i}$ axis (orthogonal to $\hat{j}$ and $\hat{k}$), R and D are the average rotational and dipole strengths, respectively, and R_{33} and D_{33} are, respectively, the rotational strength along and dipole strength polarized in the direction of the emission transition moment. R_l is given by

$$R_l = \frac{\Delta A}{2A} - \frac{2.303\Delta A \times 10^{-A}}{2(1 - 10^{-A})} \quad (2)$$

where A is the absorbance of the sample and ΔA is the absorbance for LCPL minus that for RCPL. In eq 1, if $\phi = 35.25°$, the terms in R_{33} and D_{33} vanish. Further, if only one absorbing species is present, eq 1 can be used to predict the FDCD spectrum since $4R/D$ and R_l can be obtained from transmission CD and absorption spectra.[3] Experiments on *d*-10-camphorsulfonic acid in glycerol are in agreement with the theory, but additional verification has been prevented by large artifacts.[7] These are due to imperfect CPL. For example, suppose the excitation beam contains residual linear polarization at an angle, α, to the $\hat{i}$ axis (in the $\hat{i}\hat{j}$ plane); then the fluorescence signal due to this excitation when $\phi = 0°$, $F_j(\phi = 0)$, is given by

$$F_j(\phi = 0) = F_{\parallel}\cos^2\alpha + F_{\perp}\sin^2\alpha \quad (3)$$

Here $F_{\parallel}$ and $F_{\perp}$ are the intensities of fluorescence polarized parallel and perpendicular to the excitation light, respectively. If α is different for left and right circularly polarized cycles, then a difference in fluorescence is measured that does not depend on the sample optical activity. In practice, this artifact can swamp the optical activity signal.[7] A similar artifact has plagued measurements of Raman optical activity but has recently been overcome by Hug.[9,10] We have modified Hug's detection scheme for FDCD. In the new detection system, a second photomultiplier is placed perpendicular to the first (i.e., along the $\hat{i}$ axis). The equivalent of eq 3 for this phototube is

$$F_i(\phi = 0) = F_{\parallel}\sin^2\alpha + F_{\perp}\cos^2\alpha \quad (4)$$

Summing the outputs of the two phototubes gives a signal independent of α, since $\sin^2\alpha + \cos^2\alpha = 1$. This eliminates the artifact caused only by linear polarization.[11] However, an additional artifact remains that we preliminarily attribute to finite divergence of the excitation beam. One manifestation of this artifact is that spectra with $\phi = 45°$ are not the same as spectra with no polarizer. This last artifact is removed by averaging spectra for $\pm\phi$.

The effectiveness of the two-photomultiplier system is demonstrated by the spectra of morphine in 90% glycerol shown in Figure 1. The depolarization ratio for this sample is 0.3 in the long-wavelength band,[12] and large artifacts can be observed.

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(4) Turner, D. H. *Methods Enzymol.* **1978**, *49*, 199.
(5) Ehrenberg, B.; Steinberg, I. Z. *J. Am. Chem. Soc.* **1976**, *98*, 1293.
(6) Tran, C. D.; Fendler, J. H. *J. Am. Chem. Soc.* **1979**, *101*, 1285.
(7) Lobenstine, E. W.; Turner, D. H. *J. Am. Chem. Soc.* **1979**, *101*, 2205.
(8) Tinoco, I.; Ehrenberg, B.; Steinberg, I. Z. *J. Chem. Phys.* **1977**, *66*, 916.
(9) Hug, W.; Surbeck, H. *Chem. Phys. Lett.* **1979**, *60*, 186.
(10) Hug, W. *Appl. Spectrosc.*, in press.
(11) In practice the AC($F_L - F_R$) and DC($F_L + F_R$) outputs are summed separately and then divided. The denominator is still affected by fluorescence due to linear polarization. However, this effect is small if the excitation has little residual linear polarization.

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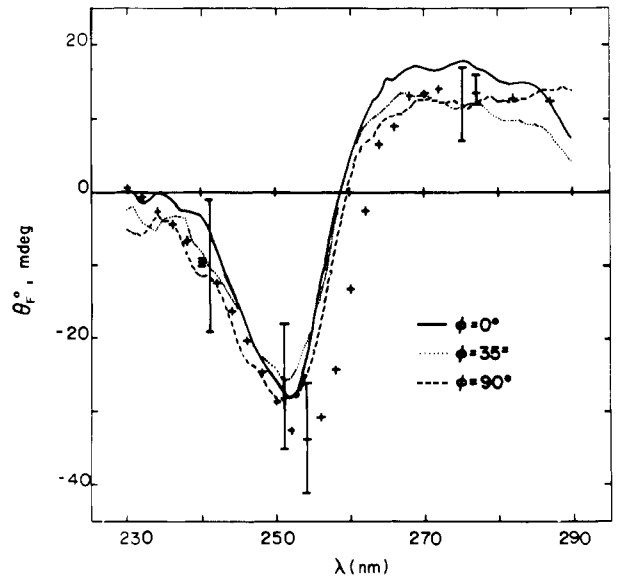

Figure 1. Photoselected FDCD spectra of 2.5×10^{-4} M morphine in 90:10 glycerol–water (v/v), 0.1 N H_2SO_4: (—) $\phi = 0°$; (···) $\phi = 35°$; (- - -) $\phi = 90°$; (+) spectrum predicted from CD and absorption data. Filter was Wratten 18-A. A cylindrical CD sample cell with 10-mm pathlength was used. Baselines were obtained by turning off the excitation modulator. Data were taken with a PDP 11/34 minicomputer; five scans each of the spectrum and baseline were averaged.

However, the average of spectra measured for $\phi = \pm 35°$ by using two phototubes agrees well with the spectrum predicted from CD and absorption measurements.[3,7] Any residual discrepancy is probably due to noise and practical problems in making the calculation. Thus the error bars for the calculated points represent the effects of 0.5-nm errors in wavelength calibration of the spectrometers.[13] Spectra for $\phi = 0°$ and 90° are also shown in Figure 1. Within experimental error, they are the same as the 35° spectrum. This implies the optical activity for excitation with k vector along the emission transition moment is similar to the average optical activity.[8]

A second potential problem for FDCD is suggested by the work of Tran and Fendler.[6,14] They report the quantum yield for L-tryptophan fluorescence differs by about 10% between excitation by LCPL and RCPL. A similar effect is observed with D-tryptophan. All current theories of FDCD assume this difference is negligible, so the measurement reflects only the difference in *absorption* for LCPL and RCPL. The disagreement is serious since current theories predict an FDCD signal of about −5 mdeg for L-tryptophan, whereas the effect described by Tran and Fendler would result in a signal of +1600 mdeg. They also point out that no FDCD spectra have been measured for enantiomers. To fill this gap, we report in Figure 2 the FDCD spectra of D- and L-tryptophan in water,[15] along with spectra predicted from CD and absorption measurements.[3] The excellent agreement and small magnitude indicate the results of Tran and Fendler are in error. We cannot rationalize this discrepancy.

The results reported here indicate the quantum yield for fluorescence is independent of the circular polarization of excitation. Therefore FDCD spectra can be related directly to transmission CD. The morphine spectra were measured for a polarization ratio and signal size similar to that expected for proteins. The good agreement with theory indicates it is now possible to measure FDCD spectra of tryptophan in proteins and interpret them much like conventional CD spectra.

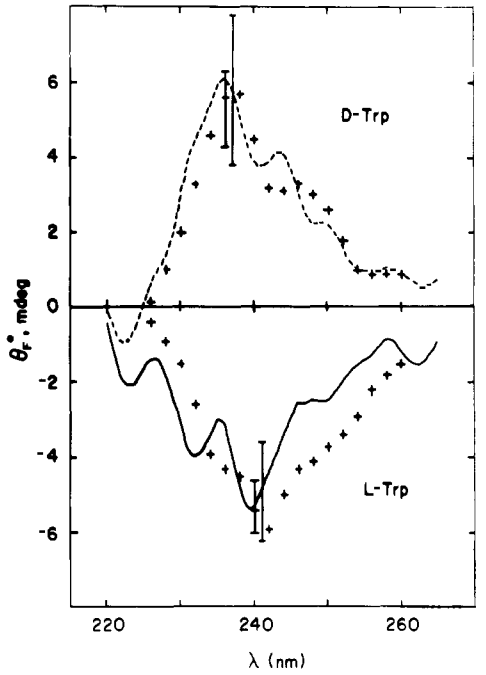

Figure 2. FDCD spectra of 5×10^{-5} M D-, L-tryptophan in water, 25 mM Na_2HPO_4, pH 10.9: (- - -), D-tryptophan; (—) L-tryptophan; (+) spectrum predicted from CD and absorption data. No polarizer was used since the fluorescence is isotropic. Baseline was 5×10^{-5} M DL-tryptophan. DL-Tryptophan and indole baselines were coincident. See Figure 1 for other conditions.

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(12) The linear depolarization ratio is defined and measured according to Parker. (C. A. Parker, "Photoluminescence of Solutions"; Elsevier, New York, 1968; pp 53, 301.)

(13) Holmium oxide glass is used to check the wavelength calibration of the absorption and CD instruments. Because the FDCD calculation is very sensitive to this wavelength calibration, the FDCD instrument is calibrated more directly. A cell containing *d*-10-camphorsulfonic acid (CSA) is placed in the excitation beam ahead of the sample cell. The sample cell is filled with optically inactive sodium fluorescein. The sodium fluorescein fluorescence is a *direct* measure of the CSA transmission CD, which is accurately known.

(14) Tran, C. D.; Fendler, J. H. In "Origins of Optical Activity in Nature", D. C. Walker, Ed.; Elsevier, Amsterdam, 1979; Chapter 5.

(15) CD and FDCD spectra for D- and L-tryptophan in methanol, the solvent used by Tran and Fendler, were too small to measure accurately.

(16) Alfred P. Sloan Fellow.

Cationic Alkylidyne–Trirhodium Cluster Complexes. Crystal Structure of $[(\eta^5\text{-}C_5H_5)_3Rh_3(\mu\text{-}CO)_2(\mu_3\text{-}CH)]PF_6$

Sir:

In recent years a number of transition-metal compounds with bridging methylene groups (μ-CH_2) have been synthesized and characterized.[1,2] Currently, attention is focused on examining

(1) (a) $(\eta^5\text{-}C_5H_5)_2Mn_2(CO)_4(CH_2)$: Herrmann, W. A.; Reiter, B.; Biersack, H. *J. Organomet. Chem.* **1975**, *97*, 245. (b) $(\eta^5\text{-}C_5H_5)_2Rh_2(CO)_2(CH_2)$: Herrmann, W. A.; Kruger, C.; Goddard, R.; Bernal, I. *Ibid.* **1977**, *140*, 73. (c) $Pt_2Cl_2(Ph_2PCH_2PPh_2)_2(CH_2)$: Brown, M. P.; Fisher, J. R.; Franklin, S. J.; Puddephatt, R. J.; Seddon, K. R. *J. Chem. Soc., Chem. Commun.* **1978**, 749. (d) $H_2Os_3(CO)_{10}(CH_2)$: Schultz, A. J.; Williams, J. M.; Calvert, R. B.; Shapley, J. R.; Stucky, G. D. *Inorg. Chem.* **1979**, *18*, 319. (e) $Ru_2(PMe_3)_6(CH_2)_3$: Hursthouse, M. B.; Jones, R. A.; Malik, K. M. A.; Wilkinson, G. *J. Am. Chem. Soc.* **1979**, *101*, 4128. (f) $(\eta^5\text{-}C_5H_4CH_3)_2Mn_2(CO)_4(CH_2)$: Creswick, M.; Bernal, I.; Herrmann, W. A. *J. Organomet. Chem.* **1979**, *172*, C39. (g) $Fe_2(CO)_8(CH_2)$: Sumner, C. E., Jr.; Riley, P. E.; Davis, R. E.; Pettit, R. *J. Am. Chem. Soc.* **1980**, *102*, 1752.

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